(19) Europäisches Patentamt
European Patent Office
Office européen des brevets

(11) EP 0 967 220 A1

(12) EUROPEAN PATENT APPLICATION

(43) Date of publication:
29.12.1999 Bulletin 1999/52

(51) Int. Cl.[6]: C07H 19/19, A61K 31/70

(21) Application number: 99111945.4

(22) Date of filing: 23.06.1999

(84) Designated Contracting States:
AT BE CH CY DE DK ES FI FR GB GR IE IT LI LU MC NL PT SE
Designated Extension States:
AL LT LV MK RO SI

(30) Priority: 24.06.1998 JP 17720998

(71) Applicant:
NIPPON ZOKI PHARMACEUTICAL CO., LTD.
Chuo-ku, Osaka (JP)

(72) Inventors:
- Yamada, Toshio,
Ono Greenery Factory,
Nippon Zoki
Minamiyama, Ono-city, Hyogo (JP)
- Yamanishi, Koichi
Toyono-gun, Osaka (JP)

(74) Representative: HOFFMANN - EITLE
Patent- und Rechtsanwälte
Arabellastrasse 4
81925 München (DE)

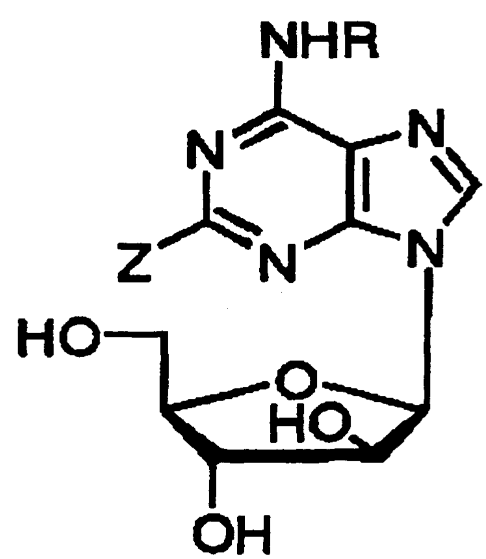
(54) **Novel arabinosyladenine derivatives**

(57) [Objects]

An object of the present invention is to offer an Ara-A derivative having a resistance to the metabolism by ADA and also having a sufficient antiviral action.

[Constitution]

A novel 2-substituted arabinosyladenine derivative represented by the formula (I) and pharmaceutically acceptable salts and hydrates thereof.



[I]

[In the formula, Z is alkyl having more than 4 carbon atoms, alkenyl or alkynyl and R is hydrogen or lower alkyl.]

EP 0 967 220 A1

[Merit]

The compounds of the present invention are Ara-A derivatives having a resistance to the metabolism by ADA and having a sufficient antiviral action whereby the problems in the prior art have been solved. The compounds of the present invention are useful as therapeutic or preventive agents for diseases infected by DNA virus such as herpes simplex virus (HSV), herpes zoster virus, cytomegalovirus (CMV), adenovirus, hepatitis virus or vaccinia virus. As compared with Ara-A, they not only show a good behavior in blood with an excellent sustaining property but also are capable of being orally administered whereby their usefulness is very high.

Description

Detailed Description of the Invention:

[Technical Field of the Invention]

[0001] The present invention relates to a 2-substituted arabinosyladenine derivative having a resistance to a metabolism by adenosinedeaminase and a pharmaceutical use thereof

[Prior Art]

[0002] Arabinosyladenine (general name: vidarabine; hereinafter referred to as „Ara-A") is effective against DNA viruses such as herpes simplex virus (HSV), herpes zoster virus, cytomegalovirus (CMV), adenovirus, hepatitis virus and vacciia virus. Clinically, it is mainly used as a therapeutic agent for infectious diseases with herpes virus. However, Ara-A is quickly metabolized by adenosinedeaminase (ADA) in blood to hypoxanthine arabinoside having a weak antiviral activity. Therefore, there is a disadvantage that its strong antiviral activity in vitro is not reflected in clinical efficacy. In addition, much ADA is present in digestive tracts and, therefore, Ara-A orally administered is metabolized before being absorbed. Accordingly, oral administration of Ara-A is so difficult and, at present, ointment and injection have only been commercially available.

[0003] Until now, various attempts have been made for stabilization of Ara-A but each of them has the following problems and is not satisfactory.

(1) A method using Ara-A and an ADA inhibitor jointly [Sloan, B., et al.: Ann. NY. Acad. Sci., vol. 284, pages 60-80 (1977)]

[0004] This is a method where Ara-A and an ADA inhibitor are simultaneously administered in order to stabilize Ara-A. Deoxycoformycin was used as an ADA inhibitor but an adverse reaction due to the combination was observed whereby the development was given up.

(2) A method making a prodrug of Ara-A [Kotra, L. P., et al.: J. Med. Chem., vol. 39, pages 5202-5207 (1996)]

[0005] This is a method where a compound in which an amino group at 6-position of Ara-A is substituted with an azide group is used as a prodrug. This azide group is reduced to an amino group by cytochrome P-450 in hepatic microsome fraction to give Ara-A in vivo. However, it is presumed that, even as compared with this reduction reaction, the metabolism by ADA is far quicker whereby it is hardly believed that the concentration of Ara-A in blood increases. Actually, although the authors described the behavior in blood of the prodrug, 6-azido-Ara-A, they did not mention at all whether the active Ara-A itself was present in blood.

(3) A method using Ara-A derivatives resistant to the metabolism by ADA [Koszalka, G. W., et al.: Antimicrobial Agents and Chemotherapy, vol. 35, pages 1437-1443 (1991); Averett, D. R., et al.: Antimicrobial Agents and Chemotherapy, vol. 35, pages 851-857 (1991)]

[0006] Syntheses of Ara-A analogs resistanct to the metabolism by ADA have been most often conducted. Koszalka et al introduced methylamino group, dimethylamino group or methoxyl group into 6-position of the base and synthesized Ara-A analogs having a resistance to the metabolism by ADA. This is also mentioned in the Japanese Laid-Open Patent Publication Sho-63/310831. However, those compounds did not show sufficient resistance to ADA. According to the investigation by the present inventors, the compound by Koszalka et al. (control compound C) into which methylamino group is introduced showed no sufficient resistance to ADA. The resistance of the compound introduced with methoxyl group was weak as well. The compound into which dimethylamino group is introduced showed a resistance to ADA. But said compound is easily demethylated to monomethyl compound in vivo and, as a result, it is also metabolized by ADA.

[0007] With regard to 2-alkyl derivatives of Ara-A, a compound where a methyl group is introduced into 2-position (control compound A) mentioned in Japanese Laid-Open Patent Publication Sho-55/45625 and a compound where an ethyl group is introduced into 2-position (control compound B) described in Keiko Sato et al. (Chem. Pharm. Bull., vol. 37, pages 1604-1608 (1989)) are listed as known substances. As a result of the test by the present inventors, no strong antiviral action was obtained as shown in Fig. 1 even when a lower alkyl group such as methyl or ethyl is introduced into 2-position of Ara-A. In addition, those compounds are not satisfactory in terms of the resistance to ADA as well.

[Problems to be Solved by the Invention]

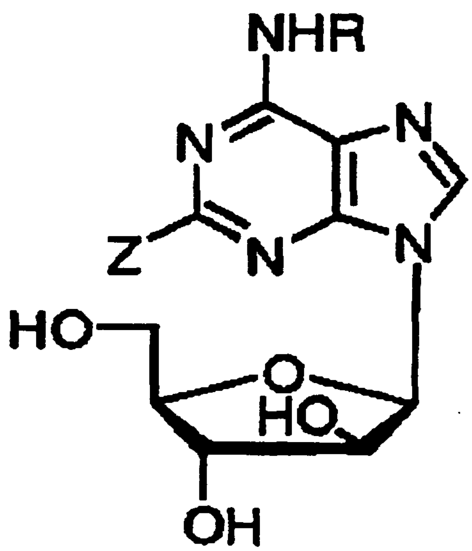
[0008] The present invention is to solve the above-mentioned problems in the prior art and is to offer an Ara-A derivative having a resistance to the metabolism by ADA and also having a sufficient antiviral action.

[Means to Solve the Problems]

[0009] The present inventors have carried out an intensive investigation on Ara-A derivatives having a high resistance to ADA and found novel 2-substituted Ara-A derivatives where the above-mentioned disadvantages are overcome. The compounds of the present invention are the compounds to which a high resistance to the metabolisms by ADA is given without deterioration of the antiviral action of Ara-A. Accordingly, the compounds of the present invention not only show good behavior in blood with an excellent sustaining property as well as Ara-A but also are capable of being applied as an oral agent. Therefore, the compounds of the present invention have very high usefulness as therapeutic or preventive agents for diseases infected by DNA virus such as herpes simplex virus (HSV), herpes zoster virus, cytomegalovirus (CMV), adenovirus, hepatitis virus or vaccinia virus.

[Best Mode for Carrying out the Invention]

[0010] The present invention relates to a 2-substituted arabinosyladenine derivative represented by the formula (I) and pharmaceutically acceptable salts and hydrates thereof Furthermore, the present invention relates to an antiviral agent containing said compounds as an effective component.



[I]

[0011] In the formula, Z is alkyl having more than 4 carbon atoms, preferred a linear or branched alkyl having 4 to 12 carbon atoms such as butyl, isobutyl, sec-butyl, tert-butyl, pentyl, isopentyl, neopentyl, hexyl, dimethylbutyl, heptyl, octyl, nonyl, decyl, undecyl or dodecyl; alkenyl, preferred a liner or branched alkenyl having 2 to 12 carbon atoms such as vinyl, propenyl, butenyl, pentenyl, hexenyl, heptenyl, octenyl, nonenyl, decenyl, undecenyl or dodecenyl; or alkynyl, preferred a liner or branched alkynyl having 2 to 12 carbon atoms such as ethynyl, propynyl, butynyl, pentynyl, hexynyl, heptynyl, octynyl, nonynyl, decynyl, undecynyl or dodecynyl. R is hydrogen or lower alkyl, preferred a linear or branched alkyl having 1 to 4 carbon atoms such as methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl or tert-butyl.

[0012] There are two asymmetric carbon atoms in the 2- and 5- position of the arabinofuranosyl ring of the compounds of formula (I) which lead to two racemic forms (±) and, therefore, four optical isomers. These racemates differ in the relative configurations of the 2- and 5-substituents which can either assume the cis- or trans-configurations. The

present invention is intended to encompass all compounds of formula (I), including the dl-or racemic mixtures as well as their separate d- and l-isomers.

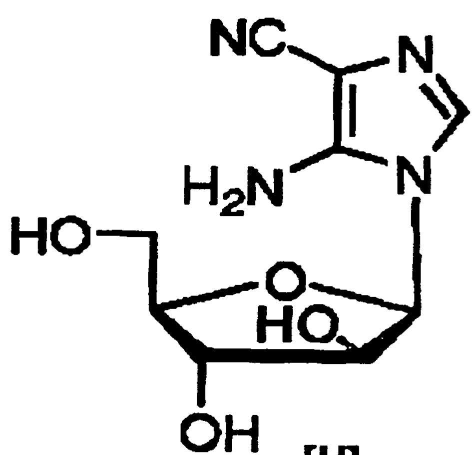
[0013] Preferred embodiments of the present invention are given as follows.

(1) A 2-substituted arabinosyladenine derivative represented by the above formula (I) and pharmaceutically acceptable salts and hydrates thereof
(2) A 2-substituted arabinosyladenine derivative according to (1) wherein Z is alkyl having more than 4 carbon atoms, alkenyl having more than 4 carbon atoms or alkynyl having more than 4 carbon atoms.
(3) A 2-substituted arabinosyladenine derivative according to (1) or (2) wherein Z is alkyl or alkynyl.
(4) A 2-substituted arabinosyladenine derivative according to (3) wherein Z is alkyl.
(5) A 2-substituted arabinosyladenine derivative according to (4) wherein Z is alkyl having 4 to 12 carbon atoms.
(6) A 2-substituted arabinosyladenine derivative according to (5) wherein R is hydrogen.
(7) A 2-substituted arabinosyladenine derivative according to (5) wherein R is alkyl.
(8) A 2-substituted arabinosyladenine derivative according to (6) wherein R is alkyl having 1 to 4 carbon atoms.
(9) A 2-substituted arabinosyladenine derivative according to (3) wherein Z is alkynyl.
(10) A 2-substituted arabinosyladenine derivative according to (9) wherein Z is alkynyl having 4 to 12 carbon atoms.
(11) A 2-substituted arabinosyladenine derivative according to (10) wherein R is hydrogen.
(12) A 2-substituted arabinosyladenine derivative according to (10) wherein R is alkyl.
(13) A 2-substituted arabinosyladenine derivative according to (12) wherein R is alkyl having 1 to 4 carbon atoms.
(14) An antiviral agent containing a 2-substituted arabinosyladenine derivative according to any of (1) to (13) as an effective component.
(15) An antiviral agent according to (14) which has a resistance to the metabolisms by adenosinedeaminase.
(16) An antiviral agent according to (14) or (15) which is a preparation for oral administration.

[0014] The above-mentioned novel Ara-A derivatives are manufactured by using the following method for example.

(Manufacturing Method No. 1)

[0015] When 5-amino-1-(β-D-arabinofuranosyl)-4-cyanoimidazole (AICN arabinoside) represented by the following formula [II] is made to react with a nitrile represented by the following formula [III], a 2-substituted Ara-A derivative represented by the above formula [I] in which R is hydrogen is obtained.



[II]

Z-CN (III)

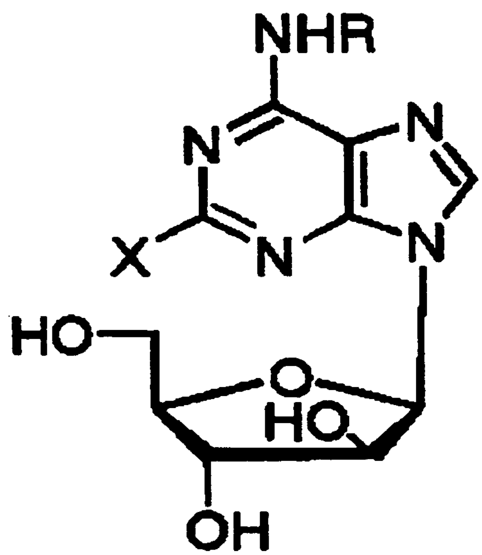
[0016] [In the formula, Z is same as that of the formula (I).]

[0017] Reaction of the compound represented by the formula [II] with the compound represented by the formula [III]

is usually carried out using from one to an excessive molar or, preferably, 1-5 mole(s) of the compound represented by the formula [III] to one mole of the compound represented by the formula [II]. Usually, a solvent saturated with ammonia gas is used in the reaction. Examples of the solvent for the reaction are alcohol such as methanol and ethanol; halogenated hydrocarbon such as methylene chloride, chloroform, carbon tetrachloride, dichloroethane and trichloroethane; ether such as ethyl ether, tetrahydrofuran and dioxane; aromatic hydrocarbon such as benzene, toluene and xylene; aprotic polar solvent such as dimethylformamide, acetonitrile and ethyl acetate; and a mixed solvent thereof and, among them, the use of alcohol is preferred. Reaction temperature is usually from room temperature to 200 °C or, preferably, from 150 °C to 200 °C. Reaction time is usually from one hour to five days or, preferably, from 1 to 24 hours.

(Manufacturing Method No. 2)

[0018] When a compound represented by the following formula [IV]



[IV]

[0019] [In the formula, R is same as that of the formula (I) and X is halogen.]
is made to react with an alkyne represented by the following formula [V],

$$R'\text{-}C{\equiv}CH \qquad [V]$$

[0020] [In the formula, R' is alkyl.]
a 2-substituted Ara-A derivative represented by the above formula [I] where Z is an alkynyl group is obtained. In that case, an alkynyl group is introduced into 2-position and it can be reduced by conventional method to give an alkyl group or an alkenyl group.

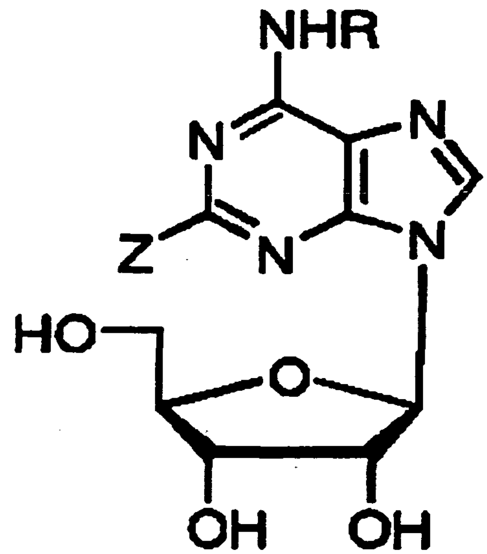
[0021] The reaction of the compound represented by the formula [IV] with the compound represented by the formula [V] is carried out using from one to an excessive molar or preferably 1.2 moles of the compound represented by the formula [V] to one mole of the compound represented by the formula [IV]. The reaction was conducted by adding 0.01-1 mole or preferably 0.1 mole of bistriphenylphosphine palladium (II), 0.5-2 mole(s) or preferably 0.5 mole of cuprous iodide and 1-5 mole(s) or preferably 1.2 moles of triethylamine to one mole of the compound represented by the formula [IV]. In conducting this reaction, it is preferred that X in the compound represented by the formula [IV] is an iodine atom. Examples of the solvent for the reaction are alcohol such as methanol or ethanol; halogenated hydrocarbon such as methylene chloride, chloroform, carbon tetrachloride, dichloroethane or trichloroethane; ether such as ethyl ether, tetrahydrofuran or dioxane; aromatic hydrocarbon such as benzene, toluene or xylene; aprotic polar solvent such as dimethylformamide, acetonitrile or ethyl acetate; and a mixed solvent thereof and, among them, the use of alcohol is

preferred. Reaction temperature is usually from room temperature to 150 °C or, preferably, from 50 °C to 100 °C. Reaction time is usually from 1 to 8 hour(s) or, preferably, from 1 to 5 hour(s).

[0022] Reduction of the 2-alkynyl derivative may be carried out by an alkali metal such as sodium or lithium or by means of a catalytic reduction using palladium carbon or Raney nickel although the use of palladium carbon is preferred.

(Manufacturing Method No. 3)

[0023] The hydroxyl groups at 3- and 5-positions of a sugar moiety of the 2-substituted adenosine derivative represented by the following formula [VI]



[VI]

[0024] [In the formula, Z and R are same as those of the formula (I).]
are appropriately protected and then a steric rearrangement reaction is carried out for 2-position of the sugar moiety followed by removing said protective group after the reaction whereupon a 2-substituted Ara-A derivative represented by the above formula [I] is obtained. Examples of the protective group are a lower alkylsilyl group such as trimethylsilyl, tert-butylsilyl and tetraisopropyldisiloxyl; a lower alkoxymethyl group such as methoxymethyl and methoxyethoxymethyl; and an aralkyl group such as trityl and, among them, a tetraisopropyldisiloxyl group is particularly preferred. The rearrangement reaction at 2-position may be carried out by a method where an alkylsulfonyl group such as a triflurometh-anesulfonyl group, a tosyl group or a mesyl group or an arylsulfonyl group is introduced as a leaving group followed by hydrolyzing, a method where a DMSO oxidation using dicyclohexylcarbodiimide and acetic anhydride as activators followed by reducing with sodium borohydride, a Mitsunobu reaction [Mitsunobu, O.: Synthesis, page 1 (1981)], etc. Removal of the leaving group may vary depending upon its type and may be carried out by a method of Green, T. W. [Protective Groups in Organic Synthesis, John Wiley & Sons (1981)] or a method similar thereto.

[0025] The compounds represented by the formulae [II], [III], [V], [VI] and [VII] used as the starting materials in the above manufacturing methods may be available from the market or may be synthesized by a method mentioned in literatures or by a method similar thereto. The compounds represented by the formulae [II] and [III] are mentioned, for example, in [Sato, U. et al.: Chem. Pharm. Bull., vol. 37, pages 1604-1608 (1989)] and [Ueeda, M. et al.: J. Med. Chem., vol. 34, pages 1334-1339 (1991)], respectively. The compound represented by the formula [IV] may be manufactured by a method where hydroxyl groups at 3- and 5-positions of the sugar moiety of 2-iodoadenosine [Nair, V., et al.: Synthesis, pages 670-672 (1982)] are appropriately protected by the above-mentioned manner, a steric rearrangement reaction for 2-position is conducted and, after the reaction, said protective group is removed.

[0026] The compound of the formula [I] obtained by the above manufacturing methods 1-3 is isolated and purified by

applying conventional separating means such as column chromatography using silica gel or adsorptive resin, liquid chromatography, solvent extraction, recrystallization or reprecipitation either solely or jointly.

[0027] The compounds represented by the above-given formula (I) include the pharmaceutically acceptable salts of thereof such as acid addition salts with hydrochloric acid, sulfuric acid, nitric acid, hydrobromic acid, phosphoric acid, perchloric acid, thiocyanic acid, boric acid, formic acid, acetic acid, haloacetic acid, propionic acid, glycolic acid, citric acid, tartaric acid, succinic acid, gluconic acid, lactic acid, malonic acid, fumaric acid, anthranilic acid, benzoic acid, cinnamic acid, p-toluenesulfonic acid, naphthalenesulfonic acid or sulfanilic acid; salts with alkali metal such as sodium or potassium, salts with alkaline-earth metal such as calcium or magnesium, and salts with other metals such as aluminum; or salts with bases such as ammonia or organic amines. Those salts may be manufactured by known methods from the compounds of the present invention in a free state or may be mutually converted among the salts.

[0028] When there are steric isomers such as cis-trans isomer, optical isomer, conformational isomer and hydrate for the substances of the present invention, the present invention includes any and all of them.

[0029] The substance of the present invention can be made into pharmaceutical preparations by a combination with a suitable pharmaceutical carriers or diluents. Any of the known methods for providing preparations, such as for oral or parenteral administrations (e.g. solids, half-solids, liquids or gases) may be used to produce the pharmaceutical compositions of the present invention. In preparing the preparations, the substance of the present invention may be used in the form of their pharmaceutically acceptable salts, and also can be used either solely or jointly together with other pharmaceutically active components.

[0030] The compounds of the present invention can be used as therapeutic or preventive agents for diseases infected by DNA virus such as herpes simplex virus (HSV), herpes zoster virus, cytomegalovirus (CMV), adenovirus, hepatitis virus or vaccinia virus, by formulating them in the suitable preparation for the administration in oral, rectal, nasal, local (including intraoral and sublingual), vaginal or parenteral (including subcutaneous, intramuscular, intracutaneous, intravenous, subarachnoidal and intradural).

[0031] In the case of preparations for oral administration, the substance of the present invention as it is or together with commonly-used excipients such as a suitable additive (e.g. lactose, mannitol, corn starch, potato starch, etc.) is mixed with binders such as crystalline cellulose, cellulose, gum arabicum, corn starch, gelatin, etc., disintegrating agents such as corn starch, potato starch, potassium carboxymethylcellulose, etc., lubricating agents such as talc, magnesium stearate, etc. and others including hulking agents, moisturizing agents, buffers, preservatives, perfumes and the like to give tablets, diluted powders, granules or capsules.

[0032] Alternatively, suppositories may be prepared by mixing with fatty/oily bases (e.g. cacao butter), emulsified bases, water-soluble bases (e.g. macrogol), hydrophilic bases, etc.

[0033] In the case of injections, it is possible to prepare the solutions or the suspensions in an aqueous and nonaqueous solvents such as distilled water for injection, physiological saline solution, Ringer's solution, plant oil, synthetic fatty acid glycerides, higher fatty acid esters, propylene glycol, etc.

[0034] In case of inhalations or aerosol preparations, the compound of the present invention in the form of a liquid or minute powder can be filled up in an aerosol container with gas or liquid spraying agent, and if desired, with conventional adjuvants such as humidifying agents or dispersing agent. They can also be used as pharmaceuticals for a non-pressurized preparation such as in a nebulizer or an atomizer.

[0035] It is also possible, depending upon the type of the disease, to prepare the pharmaceutical preparations which are other than those which were mentioned already and are suitable for the therapy such as, for example, collyriums, ointments, poultices, etc.

[0036] The preferred dose of the compound of the present invention may vary depending upon the object to be administered the patient, form of the preparation, method for the administration, term for the administration, etc. and, in order to achieve a desired effect, 0.1-100 mg per day, preferably 1-50 mg per day may be usually given to common adults by oral route.

[0037] In the case of a parenteral administration such as by injection, it is preferred that, due to the influence of the absorption, etc., a level of from 1/3 to 1/10 of the above-given dose by oral route is administered.

[0038] The present invention will be further illustrated by way of the following examples although the present invention is not limited by them at all. In the following examples, melting point was measured by placing the sample in a capillary made of glass using a melting point measuring device (type MP-21 manufactured by Yamato). Mass spectrum was measured by ionizing by means of an SIMS method using a device (type M-80B manufactured by Hitachi). Measurement of nuclear magnetic resonance (NMR) was conducted by dissolving the sample in DMSO-d_6 (containing 0.05 % of tetramethylsilane as an internal standard) using a device (ARX-500 manufactured by Brucker). Elementary analysis was conducted using a CHN corder MT-5 manufactured by Yanako.

[Examples]

Referential Example 1. Synthesis of 9-(β-D-arabinofuranosyl)-2-methyladenine [control compound A]

[0039] AICN arabinoside (1 g) and acetonitrile (2 mL) were dissolved in 50 mL of methanol saturated with ammonia of 0 °C, placed in an autoclave and heated at 140 °C for 16 hours. After completion of the reaction, the solvent was evaporated and the residue was separated and purified by a silica gel column and recrystallized from distilled water to give a control compound A (745 mg) as yellow needles.

M.P.: 247-249 °C
Mass: 282(MH^+)
^{1}H-NMR: 2.39(1H,s), 3.64(2H,m), 3.76(1H,m), 4.12(2H,m), 5.13(1H,t,j=4.9), 5.51(1H,d,j=4.4), 5.60(1H,d,j=4.9), 6.22(1H,d,j=4.9), 7.10(2H,s), 8.09(1H,s)

[0040] In the above-mentioned reaction, the nitrile corresponding to the starting acetonitrile were used and the same reaction as in Referential Example 1 was conducted to give a control compound B.

9-(β-D-arabinofuranosyl)-2-ethyladenine [control compound B]

[0041]

M.P.: 242-243 °C
Mass: 296(MH^+)
^{1}H-NMR: 1.23(3H,t,j=7.6), 2.65(2H,q,j=7.6), 3.65(2H,m), 3.76(1H,m), 4.13(2H,m), 5.10(1H,t,j-6.0), 5.52(1H,d,j=3.8), 5.62(1H,d,j=5.5), 6.24(1H,d,j=4.9), 7.09(2H,s), 8.08(1H,s)

Referential Example 2. Synthesis of 6-methylamino-9-(β-D-arabinofuranosyl)purine [control compound C]

[0042] N^6-Methyladenosine (1 g) and 1.35 g of 1,3-dichlorotetraisopropyldisiloxane were dissolved in 20 mL of pyridine and stirred at room temperature for 2 hours. After completion of the reaction, the solvent was evaporated and the residue was separated and purified by a silica gel column and recrystallized from ethyl acetate to give 1.9 g of 6-methylamino-9-[3,5-O-(tetraisopropyldisiloxan-1,3-diyl)-β-D-ribofuranosyl]purine. This compound (100 mg) was dissolved in a mixture of 4 mL of acetic anhydride and 10 mL of dimethyl sulfoxide (DMSO) and stirred at room temperature for 16 hours. After the reaction, the solvent was evaporated followed by dissolving in 20 mL of L-(-)-5,6,7,8-tetrahydrofolic acid (THF). To this solution were added 50 mg of sodium borohydride followed by stirring for 30 minutes. Then 2 mL of ethanol were added followed by stirring for 1 hour more. After small amount of acetone was added to the reaction solution, the mixture was evaporated to dryness and 1 mL of a solution of 1 M tetrabutylammonium fluoride in THF was added to the residue. The solvent was evaporated and the residue was separated and purified by a silica gel column and recrystallized from distilled water to give a control compound C (34 mg) as colorless needles.

M.P.: 194-198 °C
^{1}H-NMR: 2.95(3H,s), 3.65(2H,m), 3.78(1H,m), 4.13(2H,m), 5.09(1H,t,j=4.9), 5.51(1H,d,j=4.4), 5.61(1H,d,j=4.9), 6.26(1H,d,j=4.4), 7.70(1H,s), 8.17(1H,s), 8.22(1H,s)

Example 1.

[0043] In the reaction of Referential Example 2, the adenosine derivatives corresponding to the starting N^6-methyladenosine were used and the same reaction as in Referential Example 2 was conducted to give the compounds 1 and 2.

6-methylamino-9-(β-D-arabinofuranosyl)-2-butylpurine [compound 1]

[0044]

MP.: 165-166°C
Mass: 319(MH^+)
Elementary analysis: calculated as $C_{15}H_{23}N_5O_4 \cdot 0.2H_2O$
Theoretical: (C, 52.98; H, 6.91; N, 20.59); Found: (C, 52.98; H, 6.74; N, 20.76)

^{1}H-NMR: 0.91(3H,t,j=7.6), 1.35(2H,hex,j=7.6), 1.71(2H,qui,j=7.6), 2.67(2H,t,j=7.6), 2.95(3H,s), 3.64(2H,m), 3.76(1H,m), 4.13(2H,m), 5.09(1H,t,j=5.5), 5.51(1H,d,j=4.4), 5.61(1H,d,j=5.5), 6.24(1H,d,j=4.4), 7.50(1H,s), 8.06(1H,s)

6-methylamino-9-(β-D-arabinofuranosyl)-2-(1-butyne-1-yl)purine [compound 2]

[0045]

M.P.: 243-246 °C
Mass: 334(MH$^+$)
Elementary analysis: calculated as $C_{14}H_{22}N_6O_4$
Theoretical: (C, 49.70; H, 6.55; N, 24.84); Found: (C, 45.28; H, 6.13; N, 21.29)
^{1}H-NMR: 1.17(3H,t,j=7.6), 2.42(2H,q,j=7.6), 2.92(3H,s), 3.65(2H,m), 3.78(1H,m), 4.12(2H,m), 5.07(1H,t,j=5.5), 5,51(1H,d,j=5.5), 5.60(1H,d,j=5.5), 6.21(1H,d,j=4.9), 7.76(1H,s), 8.20(1H,s)

Example 2. Synthesis of 9-(β-D-arabinofuranosyl)-2-(1-butyne-1-yl)adenine [compound 3]

[0046]

(1) To 5 g of 2-iodoadenosine were added 50 mL of pyridine, then 4.4 g of dichlorotetraisopropyldisiloxane ($TIPDSCl_2$) were added with ice cooling and stirring and the mixture was stirred at room temperature for 1 hour. After the reaction, the solvent of the reaction solution was evaporated and the residue was recrystallized from methanol to give 2-iodo-9-[3,5-O-(tetraisopropyldisiloxan-1,3-diyl)-β-D-ribofuranosyl]-adenine (6.3 g) as colorless needles.

M.P.: 140-142 °C
^{1}H-NMR: 1.05(28H,m), 3.93(1H,dd,j=2.7, 12.5), 3.98(1H,m), 4.03(1H,dd,j=3.8, 12.5), 4.54(1H,m), 4.60(1H,dd,j=5.5, 8.7), 5.16(1H,m), 5.80(1H,s), 7.74(2H,s), 8.13(1H,s)

(2) 2-Iodo-9-[3,5-O-(tetraisopropyldisiloxan-1,3-diyl)-β-D-ribofuranosyl]adenine (970 mg) obtained in the above (1) was dissolved in 20 mL of pyridine, then 400 μL of trifluoromethanesulfonic acid chloride, 400 mg of 4-dimethylaminopyridine and 400 μL of triethylamine were added thereto with ice cooling and the mixture was stirred for 30 minutes. The reaction solution was concentrated to dryness and the residue was dissolved in 0.1 N hydrochloric acid and extracted with chloroform. The extract was separated and purified by a silica gel column. This was recrystallized from ethyl acetate to give 2-iodo-9-[2-O-trifuryl-3,5-(tetraisopropyldisiloxan-1,3-diyl)-β-D-arabinofuranosyl]-adenine (910 mg) as colorless needles. ^{1}H-NMR: 1.05(28H,m), 4.00(2H,m), 4.09(1H,m), 5.05(1H,dd,j=4.9, 8.7), 6.04(1H,d,j=4.9), 6.41(1H,s), 7.82(2H,s), 8.14(1H,s)
(3) 2-Iodo-9-[2-O-trifuryl-3,5-(tetraisopropyl-disiloxan-1,3-diyl)-β-D-arabinofuranosyl]adenine (900 mg) prepared in the above (2) was dissolved in 25 mL of N,N-dimethylformamide (DMF), 500 mg of anhydrous sodium acetate were added and the mixture was stirred at room temperature for 4 days. After completion of the reaction, the solvent was evaporated and the residue was separated and purified by a silica gel column. This was recrystallized from ethyl acetate to give 2-iodo-9-[2-O-acetyl-3,5-O-(tetraisopropyl-disiloxan-1,3-diyl)-β-D-arabinofuranosyl]adenine (633 mg) as colorless needles.

^{1}H-NMR: 1.05(28H,m), 1.67(3H,s), 3.96(2H,m), 4.16(1H,m), 4.87(1H,t,j=7.1), 5.57(1H,t,j=7.1), 6.34(1H,d,j=7.1), 7.76(2H,s), 8.00(1H,s)

(4) 2-Iodo-9-[2-O-acetyl-3,5-O-(tetraisopropyl-disioxan-1,3-diyl)-β-D-arabinofuranosyl]adenine (25 g) prepared in the above (3) was dissolved in 300 mL of THF and a solution of 1M tetrabutylammonium fluoride in 110 mL of THF was added thereto. The mixture was stirred at room temperature for 30 minutes, 100 mL of aqueous ammonia were added thereto and the mixture was stirred at room temperature for 2 hours more. After evaporation of the solvent, deionized water was added to the residue, the mixture was allowed to stand at 4 °C and the separated crystals were collected. They were recrystallized from distilled water to give 2-iodo-9-(β-D-arabinofuranosyl)adenine (13.38 g) as colorless needles.

^{1}H-NMR: 3.65(2H,m), 3.76(1H,m), 4.11(1H,m), 4.15(1H,m), 5.03(1H,t,j=5.5), 5.52(1H,d,j=4.9), 5.62(1H,d,j=4.9), 6.14(1H,d,j=5.4), 7.65(2H,s), 8.11(1H,s)

(5) 2-Iodo-9-(β-D-arabinofuranosyl)adenine (2.5 g) obtained in the above (4), 100 mg of bis(triphenylphosphine)-palladium (II) chloride, 200 mg of cupric iodide and 450 mg of butyne were dissolved in a mixture of 75 mL of DMSO and 25 mL of triethylamine and the reaction was carried out at 80 °C for 2 hours. The solvent was evaporated from the reaction solution and the residue was separated and purified by a silica gel column and recrystallized from distilled water to give the compound 3 (1.75 g) as colorless needles.

M.P.: 273-279 °C
Mass: 320(MH$^+$)
Elementary analysis: calculated as $C_{14}H_{17}N_5O_4$
Theoretical: (C, 52.66; H, 5.37; N, 21.93); Found: (C, 52.39; H, 5.28; N, 21.68)
^{1}H-NMR: 1.16(3H,t,j=7.6), 2.40(2H,q,j=7.6), 3.65(2H,m), 3.78(1H,m), 4.12(2H,m), 5.06(1H,t,j=5.5), 5.51(1H,d,j=4.4), 5.60(1H,d,j=5.4), 6.21(1H,d,j=4.9), 7.31(2H,s), 8.21(1H,s)

[0047] In the above-mentioned reaction, the alkyne corresponding to the starting butyn were used and the same reaction as in Example 2 was conducted to give the compound 4, 5 and 6.

9-(β-D-arabinofuranosyl)-2-(hexyne-1-yl)adenine [compound 4]

[0048]

^{1}H-NMR: 0.91(3H,t,j=7.1), 1.43(2H,hex,j=7.1), 1.53(2H,qui,j=7.1), 2.40(2H,t,j=7.1), 3.65(2H,m), 3.78(1H,m), 4.12(2H,m), 5.06(1H,t,j=4.9), 5.51(1H,d,j=4.9), 5.60(1H,d,j=5.5), 6.20(1H,d,j=5.5), 7.41(2H,s), 9.00(1H,s)

9-(β-D-arabinofuranosyl)-2-(octyne-1-yl)adenine [compound 5]

[0049]

^{1}H-NMR: 0.88(3H,t,j=7.1), 1.29(4H,m), 1.41(2H,m), 1.54(2H,qui,j=7.1), 2.40(2H,t,j=7.1), 3.65(2H,m), 3.78(1H,m), 4.12(2H,m), 5.06(1H,t,j=5.5), 5.51(1H,d,j=4.4), 5.60(1H,d,j=4.9), 6.20(1H,d,j=4.9), 7.31(2H,s), 8.21(2H,s)

9-(β-D-arabinofuranosyl)-2-(dodectyne-1-yl)adenine [compound 6]

[0050]

^{1}H-NMR: 0.85(3H,t,j=7.1), 1.26(16H,m), 1.40(2H,m), 1.53(2H,qui,j=7.1), 2.39(2H,t,j=7.1), 3.65(2H,m), 3.77(1H,m), 4.12(2H,m), 5.07(1H,t,j=5.5), 5.51(1H,d,j=4.4), 5.60(1H,d,j=5.5), 6.20(1H,d,j=4.9), 7.32(2H,s), 8.21(1H,s)

Example 3. Synthesis of 9-(β-D-arabinofuranosyl)-2-butyladenine [compound 7]

[0051] The compound 3 (2 g) was dissolved in 30 mL of 50 % methanol, 10 mg of 10 % palladium carbon were added thereto and the mixture was stirred at room temperature for 16 hours. The catalyst was filtered off, the filtrate was concentrated and the crystals separated out therefrom were collected. They were recrystallized from distilled water to give a compound 7 (1.95 g) as colorless needles.

M.P.: 162-163 °C
Mass: 324(MH$^+$)
Elementary analysis: calculated as $C_{14}H_{21}N_5O_4 \cdot 0.1H_2O$
Theoretical: (C, 51.72; H, 6.57; N, 21.54); Found: (C, 51.63; H, 6.49; N, 21.54)
^{1}H-NMR: 0.90(3H,t,j=7.6), 1.33(2H,hex,j=7.6), 1.69(2H,qui,j=7.6), 2.63(2H,t,j=7.6), 3.65(2H,m), 3.77(1H,m), 4.13(2H,m), 5.09(1H,t,j=6.6), 5.50(1H,d,j=4.4), 5.61(1H,d,j=5.5), 6.23(1H,d,j=4.4), 7.06(2H,s), 8.07(1H,s)

[0052] In the above-mentioned reaction, compound 4, 5 or 6 corresponding to the starting compound (compound 3) were used and the same reaction as in Example 3 was conducted to give the compound 8, 9 and 10.

9-(β-D-arabinofuranosyl)-2-hexyladenine [compound 8]

[0053]

M.P.: 98-103 °C
Mass: 352(MH$^+$)
Elementary analysis: calculated as $C_{16}H_{25}N_5O_4 \cdot 0.25H_2O$
Theoretical: (C, 54.00; H, 7.22; N, 19.68); Found: (C, 53.87; H, 7.04; N, 19.64)
^{1}H-NMR: 0.86(3H,t,j=7.1), 1.28(6H,m), 1.69(2H,qui,j=7.1), 2.62(2H,t,j=7.1), 3.64(2H,m), 3.76(1H,m), 4.12(2H,m), 5.09(1H,t,j=5.5), 5.51(1H,d,j=4.4), 5.62(1H,d,j=5.5), 6.23(1H,d,j=4.4), 7.07(2H,s), 8.07(1H,s)

9-(β-D-arabinofuranosyl)-2-octyladenine [compound 9]

[0054]

M.P.: 65-68 °C
Mass: 380(MH$^+$)
Elementary analysis: calculated as $C_{18}H_{29}N_5O_4 \cdot 0.5H_2O$
Theoretical: (C, 55.65; H, 7.78; N, 18.03); Found: (C, 55.59; H, 7.63; N, 18.19)
^{1}H-NMR: 0.86(3H,t,j=7.1), 1.27(10H,m), 1.69(2H,qui,j=7.1), 2.62(2H,t,j=7.1), 3.64(2H,m), 3.77(1H,m), 4.13(2H,m), 5.09(1H,t,j=4.9), 5.50(1H,d,j=4.4), 5.61(1H,d,j=5.5), 6.23(1H,d,j=4.9), 7.06(2H,s), 8.07(1H,s)

9-(β-D-arabinofuranosyl)-2-dodecyladenine [compound 10]

[0055]

M.P.: 67-74 °C
Mass: 436(MH$^+$)
Elementary analysis: calculated as $C_{22}H_{37}N_5O_4 \cdot 0.45H_2O$
Theoretical: (C, 59.56; H, 8.61; N, 15.78); Found: (59.49; H, 8.54; N, 15.85)
^{1}H-NMR: 0.85(3H,t,j=7.1), 1.25(18H,m), 1.67(2H,m), 2.61(2H,t,j=7.1), 3.65(2H,m), 3.76(1H,m), 4.13(2H,m), 5.10(1H,t,j=5.5), 5.51(1H,d,j=4.9), 5.61(1H,d,j=5.5), 6.23(1H,d,j=4.9), 7.07(2H,s), 8.07(2H,s)

Example 4. Measurement of antiviral activity.

[0056] Antiviral activity of the compound of the present invention was investigated by the following pharmacological experiments.

[0057] As a test for efficacy as antiviral activity, antiviral activity to shingles herpes virus was measured. Cell-free aricella-zoster virus (VZV) (Kawaguchi strain) was diluted and the resulting solution was layered on HEL cells of a 100% confluent. This was placed in a CO_2 incubator of 37 °C, shaken every 15 minutes and infected for one hour. The virus solution was removed by suction, then DMEM containing 5 % of FCS and a compound of the present invention, a control compound or Ara-A (positive control) was added thereto and the mixture was incubated for one week. This was fixed by formalin, dyed with Crystal Violet and numbers of plaques were measured. The inhibiting rate to shingles virus was determined from the plaque numbers of the control. The result to shingles virus is shown in Fig. 1 and Fig. 2. The compounds of the present invention had a sufficient antiviral activity to shingles virus. Further, the antiviral activity of the compounds of the present invention to herpes simplex virus type 1 and type 2 was measured by the same manner as mentioned above and, as a result, the compounds of the present invention had a sufficient antiviral activity to herpes simplex virus type 1 and type 2 as well.

Example 5. Measurement of stability to ADA.

[0058] Stability of the compounds of the present invention to ADA was investigated by the following biochemical experiments.

[0059] An enzymatic reaction was carried out under the following conditions. Thus, 200 µL (final concentration: 0.1 mM) of a 1 mM solution of the compound of the present invention, a control compound or Ara-A (control), 1.5 mL of a 0.1 M phosphate buffer (pH 7.5; final concentration: 75 mM) and 300 µL of a 200 units/mL of enzyme solution (final concentration: 20 units/mL) were placed in a UV cell and a decrease in ultraviolet absorption at 265 nm with a lapse of time was measured by an ultraviolet absorption meter. The enzymatic reaction was carried out at 25 °C. As the enzymatic

reaction proceeded, an ultraviolet absorption at 265 nm decreased and said rate of decrease in ultraviolet absorption was expressed as a relative enzymatic reaction rate. An example of the result is shown in Fig. 4. The control compound C had no sufficient resistance to the metabolism by ADA while all of the compounds of the present invention were very stable to the metabolism by ADA.

[Merit of the Invention]

[0060] As shown in Fig. 1 and Fig. 2, a sufficient antiviral action is not achieved even when a lower alkyl group such as methyl or ethyl is introduced into 2-position of Ara-A (control compounds A and B). However, the compounds of the present invention where an aliphatic hydrocarbon group having 4 or more carbons is introduced into 2-position of Ara-A showed the good antiviral action as same as Ara-A.

[0061] In addition, as shown in Fig. 3, the compound where a substituent is introduced into 6-position of the base portion of Ara-A (control compound C) was metabolized by ADA. Further, although not shown in the drawing, the compounds where methyl or ethyl group is introduced into 2-position of Ara-A (control compounds A and B) did not show a favorable resistance to ADA.

[0062] On the contrary, regarding the compounds of the present invention where an aliphatic hydrocarbon group having 4 or more carbons is introduced into 2-position of Ara-A, the decomposition was completely inhibited and had a sufficient resistance to the metabolism by ADA. Therefore, the oral administration of the known compounds as said control compounds have been impossible, however, the compounds of the present invention can be applicable to oral administration.

[0063] As such, the compounds of the present invention are Ara-A derivatives having a resistance to the metabolism by ADA and having a sufficient antiviral action whereby the problems in the prior art have been solved. The compounds of the present invention are useful as therapeutic or preventive agents for diseases infected by DNA virus such as herpes simplex virus (HSV), herpes zoster virus, cytomegalovirus (CMV), adenovirus, hepatitis virus or vaccinia virus. As compared with Ara-A, they not only show a good behavior in blood with an excellent sustaining property but also are capable of being orally administered whereby their usefulness is very high.

<u>Brief Explanation of the Drawings</u>:

[0064]

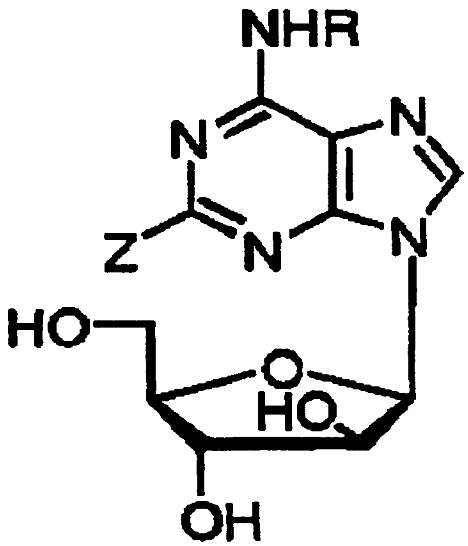
Fig. 1 is an example of the result showing the inhibitory activity of the compounds of the present invention against herpes zoster virus.

Fig. 2 is also an example of the result showing the inhibitory activity of the compounds of the present invention against herpes zoster virus.

Fig. 3 is an example of the result showing a resistance of the compound of the present invention to the metabolism by adenosinedeaminase (ADA).

Claims

1. A 2-substituted arabinosyladenine derivative represented by the following formula (I):



[I]

wherein Z is an alkyl group having more than 4 carbon atoms, an alkenyl group or an alkynyl group, and R is a hydrogen atom or a lower alkyl group, and pharmaceutically acceptable salts and hydrates thereof.

2. A pharmaceutical composition comprising a 2-substituted arabinosyladenine derivative as defined in claim 1 or a pharmaceutically acceptable salt or hydrate thereof.

3. The pharmaceutical composition according to claim 2, which is suitable for oral administration.

4. Use of a 2-substituted arabinosyladenine derivative as defined in claim 1 or a pharmaceutically acceptable salt or hydrate thereof for the preparation of a medicament which is useful as an antiviral agent.

5. The use according to claim 4, wherein the medicament is suitable for oral administration.

Fig. 1

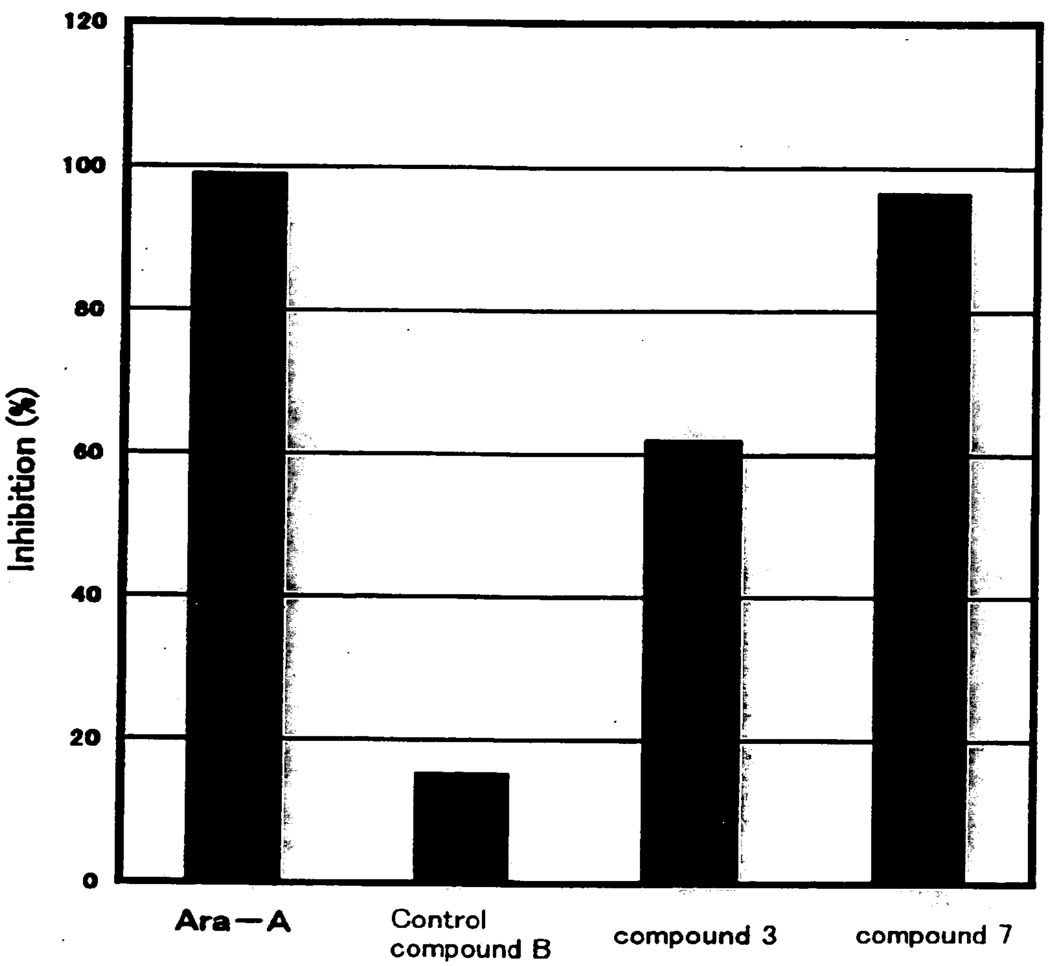
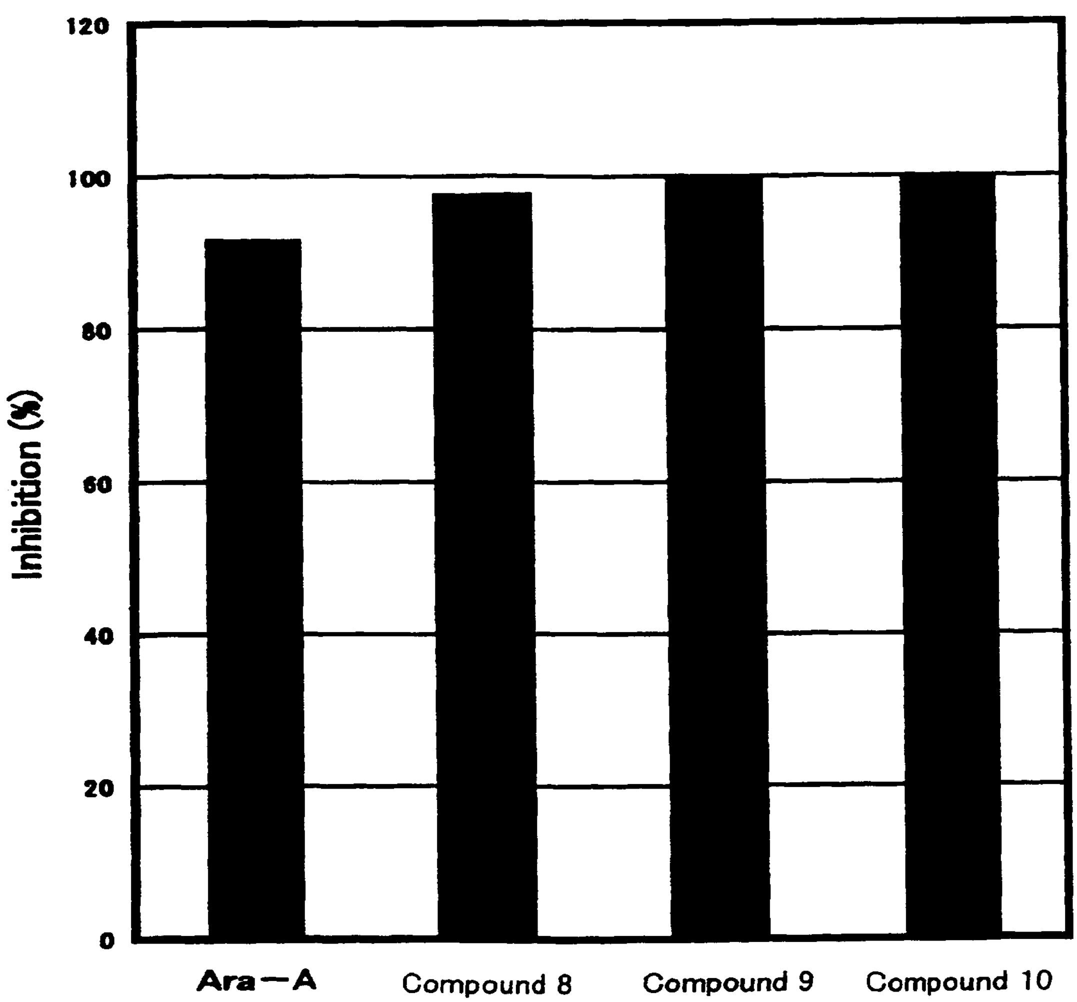
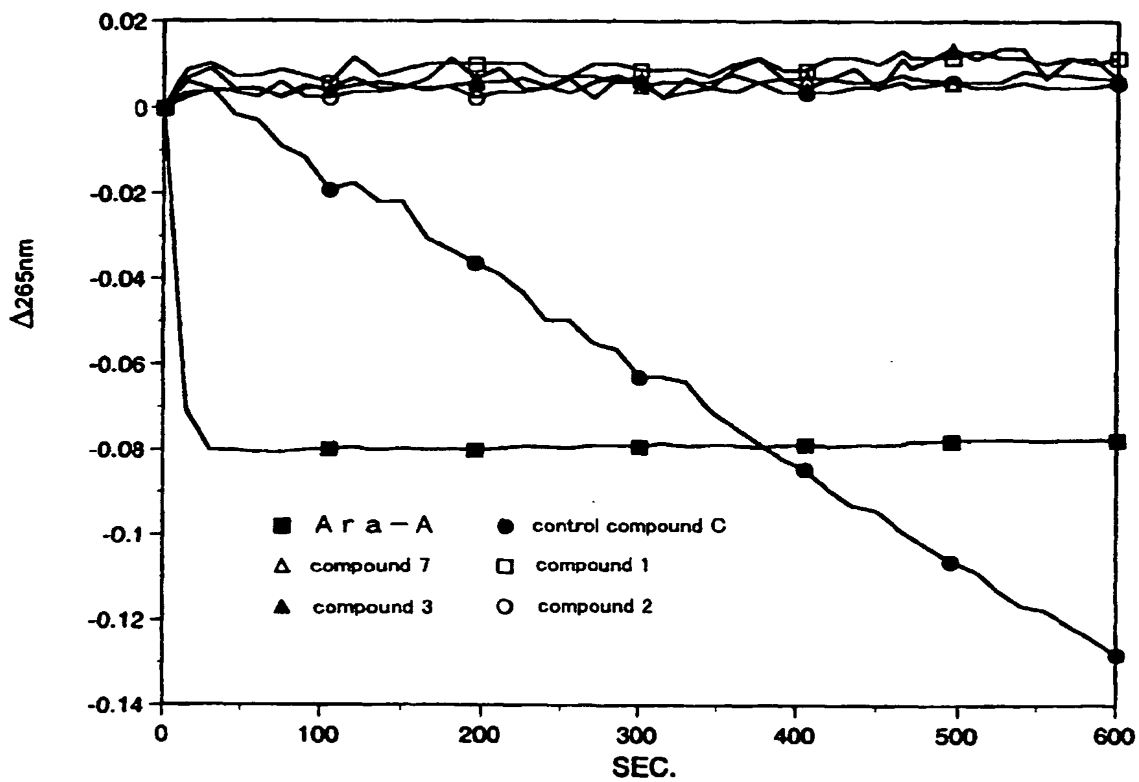

120
100
80
60
40
20
0
Inhibition (%)
Ara—A
Control compound B
compound 3
compound 7

Fig. 2



120
100
80
60
40
20
0
Inhibition (%)
Ara—A
Compound 8
Compound 9
Compound 10

Fig. 3


0.02
0
-0.02
-0.04
-0.06
-0.08
-0.1
-0.12
-0.14
Δ265nm
0
100
200
300
400
500
600
SEC.
Ara−A
control compound C
compound 7
compound 1
compound 3
compound 2

European Patent Office

EUROPEAN SEARCH REPORT

Application Number
EP 99 11 1945

DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (Int.Cl.6)
D,A	Y.SATO ET AL.: "Synthesis of 2-Substituted Adenine-arabinosides and Related Compounds from 5-Amino-4-Cyano-1-B-D-ribofuranosyl)imidazole." CHEMICAL AND PHARMACEUTICAL BULLETIN., vol. 37, no. 6, 1989, pages 1604-1608, XP002117048 PHARMACEUTICAL SOCIETY OF JAPAN. TOKYO., JP ISSN: 0009-2363 * the whole document * ---	1,2,4	C07H19/19 A61K31/70
D,A	CHEMICAL ABSTRACTS, vol. 95, no. 5, 3 August 1981 (1981-08-03) Columbus, Ohio, US; abstract no. 43582, AJINOMOTO CO. INC.: "Purine Nucleosides." page 809; column 1; XP002117049 * abstract * & JP 55 160797 A (AJINOMOTO CO. INC.) 13 December 1980 (1980-12-13) -----	1,2,4	TECHNICAL FIELDS SEARCHED (Int.Cl.6) C07H

The present search report has been drawn up for all claims

Place of search	Date of completion of the search	Examiner
THE HAGUE	30 September 1999	Scott, J

CATEGORY OF CITED DOCUMENTS

X : particularly relevant if taken alone
Y : particularly relevant if combined with another document of the same category
A : technological background
O : non-written disclosure
P : intermediate document

T : theory or principle underlying the invention
E : earlier patent document, but published on, or after the filing date
D : document cited in the application
L : document cited for other reasons

& : member of the same patent family, corresponding document

EPO FORM 1503 03.82 (P04C01)

ANNEX TO THE EUROPEAN SEARCH REPORT ON EUROPEAN PATENT APPLICATION NO. EP 99 11 1945

This annex lists the patent family members relating to the patent documents cited in the above-mentioned European search report.
The members are as contained in the European Patent Office EDP file on
The European Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

30-09-1999

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
JP 55160797 A	13-12-1980	NONE	

EPO FORM P0459

For more details about this annex : see Official Journal of the European Patent Office, No. 12/82

THIS PAGE BLANK (USPTO)